

**REMARKS**

The Office Action dated January 14, 2003, presents the examination of claims 1-14. Claims 8, 10-12, and 14 are canceled. Claims 1-4, 6, 9, and 13 are amended. No new matter is inserted into the application.

***Request for Interview***

If the instant Reply after Final for any reason does not place the present application into condition for allowance, the Examiner is respectfully requested to contact Kristi L. Rupert, Ph.D. (Reg. No. 45,702) at 703-205-8000, to schedule a personal interview prior to the issuance of an Advisory Action. }

***Rejection under 35 U.S.C. § 112, Second Paragraph***

Claims 1-14 stand rejected under 35 U.S.C. § 112, second paragraph, for allegedly being indefinite. Claims 8, 10-12, and 14 are canceled, thus rendering rejection of these claims moot. Applicants respectfully traverse the rejection applied to the pending claims. Reconsideration of the claims and withdrawal of the instant rejection are respectfully requested.

**Claim 1**

The Examiner asserts that the phrase "Agrobacterium based

plant transformation" is unclear. In response to the Examiner's remarks, Applicants amend the phrase to "*Agrobacterium*-mediated plant transformation." This phrase is well known and used in the art, as evidenced by Martineau et al., *The Plant Cell*, 6(8):1032-1033 (1994), a copy of which is attached hereto as **Exhibit 1**. Martineau utilizes a similar expression, "*Agrobacterium*-mediated DNA transfer system."

The principle of *Agrobacterium*-mediated plant transformation is described in the specification (see page 1, line 16 to page 2, line 11). The skilled artisan will recognize that *Agrobacterium*-mediated plant transformation is performed basically by the following steps:

(i) preparing a transformation vector comprising in the T-DNA region, a foreign gene to be integrated into the genome of the plant cell,

(ii) preparing *Agrobacterium* containing the above-prepared transformation vector, and

(iii) infecting the plant cell with the above-prepared *Agrobacterium* to integrate the foreign gene into the plant genome.

For these reasons, Applicants respectfully submit that the phrase "*Agrobacterium*-mediated plant transformation" distinctly claims the subject matter of the present invention such that the

requirements of 35 U.S.C. § 112, second paragraph are met. Withdrawal of the instant rejection is therefore respectfully requested.

Claim 2

The Examiner rejects claim 2 for the recitation of "recognized," which the Examiner asserts is unclear. Applicants respectfully disagree. The term "recognize" is known to the skilled artisan as a common word used to refer to the function of a protein. Generally speaking, when a protein acts in a site-specific manner, there are mechanisms in which the protein actually identifies and acts on the site. Such mechanisms are referred to by the term "recognize."

According to Sheng et al., *vir* proteins are known in the art to "function together as an endonuclease that carries out site- and strand-specific nicks between the third and fourth base pair of the bottom strand of the T-DNA borders." See, page 1703, left column, lines 18-21 of Sheng et al., *The Plant Cell*, 8:1899-1710 (1996), attached hereto as **Exhibit 2**. This means that the *vir* proteins actually bind specifically to T-DNA border sequences and act as an endonuclease. In other words, the *vir* proteins "recognize" T-DNA border sequences and exert their function thereon.

For these reasons, Applicants respectfully submit that the

term "recognize" distinctly claims the subject matter of the present invention such that the requirements of 35 U.S.C. § 112, second paragraph are met. Withdrawal of the instant rejection is therefore respectfully requested.

Claims 5 and 6

The Examiner notes that the marked-up copy of claims 5 and 6 does not match the clean copy of the claims. Specifically, the Examiner states that "Agrobacterium" is misspelled in the claims. Regarding claim 5, Applicants respectfully submit that "Agrobacterium" is correctly spelled therein and has always been so spelled. Regarding claim 6, Applicants submit two versions: one as twice amended in the Reply filed on October 23, 2002, and a newly three-times amended version. In the twice-amended version, the misspelled "Angorbacterium" is deleted. In the thrice-amended version, all recitations of "Agrobacterium" are correctly spelled and the clean copy thereof matches the marked-up copy. Thus, the instant rejection is overcome.

Claims 6 and 9

The Examiner asserts that claims 6 and 9 are incomplete method claims. In response to the Examiner's remarks, Applicants amend

the claims so that a transformed plant is produced by the final step of the method. Thus, the instant rejection is overcome.

Applicants respectfully submit that the above remarks and/or amendments address and overcome the rejection of the claims under 35 U.S.C. § 112, second paragraph. Withdrawal thereof is therefore respectfully requested.

***Rejection under 35 U.S.C. § 112, First Paragraph***

**Enablement**

Claims 1-14 are rejected under 35 U.S.C. § 112, first paragraph, for allegedly not being enabled by the specification. Claims 8, 10-12, and 14 are canceled, thus rendering rejection of these claims moot. Applicants respectfully traverse the rejection applied to the pending claims. Reconsideration of the claims and withdrawal of the instant rejection are respectfully requested.

The Examiner maintains her assertion that the specification does not enable "all" modifications of the left border sequence, nor a vector which "prevents" integration of a non-T-DNA segment into a plant chromosome.

Claim 1, as amended, recites a T-DNA left border region comprising more than one T-DNA left border sequence. It is noted

that "more than one" includes not only whole numbers but also decimal fractions.

One of the embodiments of the present invention utilizes more than one T-DNA left border sequence as the T-DNA left border region, in order to ensure that the *vir* proteins recognize the T-DNA left border regions and to reduce the probability of integration of non-T-DNA segment into transformed plant genomes. In the Examples provided in the specification, the T-DNA left border region is made up of two left border sequences. The employment of this feature in *Agrobacterium*-mediated plant transformation results in reduced integration of any non-T-DNA segment, when compared with the situation in which a T-DNA left border region with a single left border sequence is employed. Further, a T-DNA left border region with three left border sequences reduces undesired integration even more. These data demonstrate that the greater the number of the T-DNA left border sequences, the greater the effect of reducing integration of any non-T-DNA segment.

Turning to the "preventing" part of the Examiner's rejection, Applicants amend the claims to change "prevent" to "reduces integration frequency." As acknowledged by the Examiner, the specification teaches a reduction in integration frequency of non-

T-DNA sequences.

Applicants respectfully submit that the claims, as amended, fully meet the requirements of 35 U.S.C. § 112, first paragraph. Withdrawal of the instant rejection is requested.

New Matter

Claims 1-14 are rejected under 35 U.S.C. § 112, first paragraph, for allegedly containing new matter. Claims 8, 10-12, and 14 are canceled, thus rendering rejection of these claims moot. Applicants respectfully traverse the rejection applied to the pending claims. Reconsideration of the claims and withdrawal of the instant rejection are respectfully requested.

The Examiner asserts that the phrase "prevents integration of a non-T DNA segment into a plant chromosome" is new matter. Applicants amend the phrase to "reduces integration frequency of a non-T-DNA segment into a plant chromosome."

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The Examiner also asserts that the amendment of "marker gene" to "marker" in claim 4 is improper because there is allegedly no support for "marker" in the specification. Applicants amend the phrase back to "marker gene".

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Applicants respectfully submit that the claims, as amended, contain no new matter. Withdrawal of the instant rejection is therefore respectfully requested.

**Rejection under 35 U.S.C. § 102**

The Examiner rejects claims 1, 2, 4-10 and 14 under 35 U.S.C. § 102(b) for allegedly being anticipated by Becker et al. (of record). Claims 8, 10, and 14 are canceled, thus rendering rejection of these claims moot. Applicants respectfully traverse the rejection applied to the pending claims. Reconsideration of the claims and withdrawal of the instant rejection are respectfully requested.

The Examiner asserts that the vector of Baker et al. must possess a modified left border region since there is no evidence for integration of any non-T-DNA segment in the plant chromosomes. Applicants respectfully disagree. The true reason why integration is not mentioned in Baker et al. is that Baker et al. simply did not know of, nor recognize, the problem of non-T-DNA integration.

Martineau et al. (**Exhibit 1**) describes that the general view in the art in 1992 was that no DNA segment outside of the T-DNA border is transferred into plant genomes. However, Martineau et al. teaches in the publication that some DNA segments located outside the T-DNA border are integrated into plant genomes in approximately 20 to 30% of transformed plants.

Therefore, at the time of publication of Baker et al. in 1992, the general view held in the art was that non-T-DNA segment integration did not occur. For this reason, Baker et al. did not



have any motivation to reduce integration of any non-T-DNA segments, and certainly did not have any motivation to investigate the presence of such an integration. After publication of Baker et al., the integration of the non-T-DNA segment came to be noticed as a serious problem in the field of plant transformation. Thus, the object of the present invention is to solve this problem; namely, to reduce non-T-DNA segment integration. Therefore, the invention of Baker et al. and the present invention are distinguishable.

In addition, the instant claims are amended so that the phrase "modified T-DNA left border region" is replaced with "T-DNA left border region comprising more than one T-DNA left border sequence."

Thus, the present invention is completely distinguishable from Becker et al. Withdrawal of the instant rejection is respectfully requested.

### **Conclusion**

All of the stated grounds of rejection have been properly traversed, accommodated, or rendered moot. Applicants therefore respectfully request that the Examiner reconsider all presently outstanding rejections and that they be withdrawn. It is believed that a full and complete response has been made to the outstanding

Office Action and, as such, the present application is in condition for allowance.

Pursuant to the provisions of 37 C.F.R. §§ 1.17 and 1.136(a), the Applicants hereby petition for an extension of two (2) months to June 14, 2003, in which to file a reply to the Office Action. The required fee of \$410.00 is enclosed herewith.

If necessary, the Commissioner is hereby authorized in this, concurrent, and future replies, to charge payment or credit any overpayment to Deposit Account No. 02-2448 for any additional fees required under 37 C.F.R. §§ 1.16 or 1.17; particularly, extension of time fees.

Respectfully submitted,

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Attachments: Version with Markings to Show Changes Made;  
**Exhibit 1:** Martineau et al., *The Plant Cell*,  
6(8):1032-1033 (1994);  
**Exhibit 2:** Sheng et al., *The Plant Cell*, 8:1899-  
1710 (1996).

VERSION WITH MARKINGS TO SHOW CHANGES MADE

IN THE CLAIMS:

Claims 8, 10, 11, 12, and 14 are canceled.

The claims have been amended as follows:

1. (Twice Amended) A vector for Agrobacterium-mediated  
[Agrobacterium based] plant transformation, comprising

a [modified] T-DNA left border region comprising more than one  
T-DNA left border sequence [,

wherein said vector prevents integration of a non-T-DNA  
segment into a plant chromosome].

2. (Twice Amended) A vector for Agrobacterium-mediated  
[Agrobacterium based] plant transformation comprising:

a T-DNA right border region that is recognized by the vir  
proteins of Agrobacterium;

a [modified] T-DNA left border region comprising more than one  
T-DNA left border sequence that is recognized by the vir proteins  
of Agrobacterium;

a T-DNA region located between these border regions and into  
which a nucleotide sequence to be introduced into the plant can be  
inserted; and

a replication origin that enables replication of said vector in bacteria,

wherein said vector reduces the integration frequency of a [prevents integration of any] non-T-DNA segment into a plant chromosome, as compared with a vector comprising a T-DNA left border region consisting of a single T-DNA left border sequence, when said vector is used in the Agrobacterium-mediated plant transformation.

3. (Twice Amended) The vector according to claim [1 or] 2, wherein the [modified] T-DNA left border region comprises at least two [a plurality of] T-DNA left border sequences.

4. (Three Times Amended) The vector according to claim 2, wherein the T-DNA region contains a marker gene that permits the selection of a plant transformed with the vector.

6. (As Twice Amended in the Reply filed on October 23, 2002)  
A method [of] for transforming [plants] a plant comprising the steps of:

[using an *Agrobacterium* host cell containing] introducing the vector according to [claim 1] any one of claims 1, 2, 4 or 5 into an *Agrobacterium* host cell; and  
transforming a plant cell with the *Agrobacterium* host cell harboring the vector.

6. (Three Times Amended) A method for transforming a plant comprising the steps of:

introducing the vector according to any one of claims 1, 2, 4 or 5 into an *Agrobacterium* host cell; and

transforming a plant cell with the *Agrobacterium* host cell harboring the vector,

thus obtaining a transformed plant.

9. (Amended) A method for reducing the integration frequency [preventing integration] of non-T-DNA segment of a vector for *Agrobacterium*-mediated [*Agrobacterium* based] plant transformation, comprising the steps of:

introducing the vector according to any one of claims 1, 2, 4 or 5 into an *Agrobacterium* host cell; and

transforming a plant cell with the *Agrobacterium* host cell harboring the vector,

thus obtaining a transformed plant cell, wherein the integration frequency of non-T-DNA segment into the chromosome of the plant cell is reduced as compared to the case when a vector comprising a T-DNA left border region consisting of a single T-DNA left border sequence is used.

13. (Amended) The vector according to claim 2 [3], wherein the [modified] T-DNA left border region comprises at least [more than] three [repeats of the] T-DNA left border sequences [sequence].